# IMPROVEMENT OF CONCEPTION RATE IN CROSSBRED CATTLE BY USING GNRH ANALOGUE THERAPY

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#### Abstract

The study was carried out to evaluate the effect of Dalmarelin (Lecirelin acetate, GnRH analogue) administration at the time of AI on conception rate in repeat breeding crossbred cattle. A total of 64 cows maintained at the Government Farm Okara were included in the study. Age of these cows ranged from 44 to 232 months and their lactation number varied from 1-13. The experimental cows were divided into control and treatment groups. The average age and lactation number of cows of the control and treatment groups did not differ. In this experiment 32 cows received no treatment at the time of AI while in other group 32 cows received Dalmarelin (GnRH analogue) therapy at the time of AI. The cows were examined after 60 days post insemination per rectum. A total 12 animals out of 32 confirmed pregnant after rectal examination having conception rate 68.75% (P < 0.0242/x2: 5.082). The data regarding postpartum interval, estrus duration and estrus behaviour was collected and analyzed, which was non-significant. The study revealed that use of GnRH analogue therapy improved the conception rate in repeat breeding crossbred cows when it was administered at the time of AI.

Key words: conception rate, repeat breeding, crossbreed cows, GnRH analogy therapy

#### **INTRODUCTION**

Pakistan is rich in livestock wealth. According to 2006 census, there were 29.56 million cattle and 27.34 million buffalo in the country (Anonymous, 2006). Livestock population in the country is scattered i.e. there is little organized livestock farming and animals are kept in smallholding of 1 to 10 per owner. The average milk yield of cattle and buffalo was 4.5 litter and 5.5 litters, respectively. This yield was remarkably low as compared to average milk yield per animals in developed countries. The main reason of low milk yield of our livestock is due their inferior genetic makeup. To increase the milk yield per animals, it is necessary to improve the genetic makeup of our livestock through genetic improvement using technologies like AI and Embryo Transfer.

Crossbreeding provides an important tool for improving the production potential of non-descript cattle by crossing with higher milk producing exotic dairy breed semen. Crossbreeding has resulted in reduction of 10–12 months in age of maturity at first calving. Milk production in crossbred animals has also improved by 75–140 percent over their indigenous parents (Anonymous, 1996). Presently, a fair number of crossbred cattle are found in the country.

Gonadotropin releasing hormone (GnRH) is a protein produced by hypothalamus. This hormone controls the synthesis and release of luteininzing hormone (LH) and follicle stimulating hormone (FSH), both originating from the pituitary gland. The combined action of these two hormones regulates follicular development, ovulation and corpus luteum (CL) function. For many years, these GnRH agonists have been used to manipulate reproduction functions in dairy cattle (Douglas, 1998). Various GnRH agonists have been used with variable success for enhancing pregnancy rates and shortening the interval to first postpartum ovulation in dairy cattle. The different economical and more reliable methods are used to evaluate financial returns from the use of GnRH at time of insemination to enhance fertility of dairy cows. Some studies have shown that response to GnRH treatment varied with the herd fertility level. Herds with conception rate 60% benefited from the treatment only at second or later services. Selection of the second / third insemination for the GnRH as treatment services usually resulted in the greatest total return (Weaver et al., 1988). The enhancement of fertility necessary to achieve the break-even point with GnRH treatment at third service was 2% for low and 5% for higher conception rate herds. Baseline herd conception rate, estrus detection efficiency, replacement cost, value of excessive days not pregnant and cost of treatment has the greatest effect on return from treatment. Herds with higher conception rate and low replacement cost are likely to get least benefit from GnRH treatment at insemination is profitable under most herd condition.

This study was designed to examine the potential use of exogenous GnRH given at the time of insemination for controlling the incidence of repeat breeding in crossbred dairy cattle.

# MATERIALS AND METHODS

# 1. Animal Selection

The study was conducted at Government Farm Okara from March to May 2004. A total of 64 healthy cyclic crossbred (Friesian × Sahiwal and Red Sindhi) repeat breeder cows were selected randomly for the present project. Age of these cows ranged from 44 to 232 months and their lactation number varied from 1–13. These 64 selected experimental crossbred cows were divided into two groups comprising of 32 cows in each group i.e. Group 'A' was kept as control and Group B was treatment group which received treatment with Dalmarelin. Normalcy of genitalia of all the animals was confirmed through rectal palpation at the start of the studies.

### 2. Management

These all 64 animals were maintained under same routine feeding and management conditions of the farm. The routine feeding consisted of available green fodder plus concentrate ration according to the daily milk yield. Vaccination was carried out as per vaccination schedule.

### 3. Heat detection

- 3.1 Estrus detection of cows was carried out intensively twice daily (0600 to 0900 hrs am and pm) by experienced herd persons. Only those animals were selected and recorded in heat which stood still while being mounted/ridden by other female cows and vasectomized bulls.
- 3.2 The animals were also observed for behavioural symptoms like frequent urination, bellowing, raised tail, restlessness and licking of external genitalia.
- 3.3 Different visible external changes like vulvular oedema and absence of wrinkles on vulvular lips, vaginal hyperaemia, wetness and mucus discharge were also observed.

### 4. Treatment

The crossbred cows of Group 'A' served as control as no treatment of Dalmarelin was given to them at the time of insemination. Group B, comprising of crossbred cows, received the treatment with Dalmarelin.

*4.1 Dose Rate.* As recommended by manufactures single injection of Dalmarelin (50 ug Lecirelin) 2 ml was injected intramuscularly at the time of insemination. *4.2 Treatment schedule* 

Treatment	Group 'A'	Group 'B'
No of animals	32	32
Dalmarelin (50 ug Lacirelin) given I/M at the time of insemination	No Treatment	Treatment Given

### 5. Artificial Insemination by Frozen Thawed Semen

Frozen semen from  $3 \times$  Friesian bulls was used and evenly allotted to the cows in both the groups. The semen was stored in liquid nitrogen at temperature of  $-196^{\circ}$ C. The semen was thawed at temperature  $37-40^{\circ}$ C in water bath for 10–15 sec and after washing the external genitalia with water the cows were inseminated. Sterilized Insemination Gun with disposable plastic sheath was used for insemination of the entire animal. All the hygienic measures were adopted to check the possibility of infection at the time of artificial insemination.

#### 6. Experimental period

The duration of study was three months from the start of experiment (from March to May 2004).

### 7. Duration of estrus

Time from the onset of first sign of estrus to end of estrus was recorded for each cow in hours.

#### 8. Parameters observed in the study

Following parameters were recorded for individual animal before the start of the study.

- 8.1 Age and lactation number.
- 8.2 Duration of estrus and estrus behaviour (Weak, Moderate, Intense).
- 8.3 The estrus behaviour was scored and judged using under mentioned criteria.
  - a. Weak estrus behaviour. Animals showing heat signs for a period of 7–10 hours.
  - b. Moderate estrus behaviour. Animals showing heat signs for a period of 11–16 hours.
  - c. Intense estrus behaviour. Animals showing heat signs for a period of 17–24 hours.
- 8.4 Symptoms of estrus
- 8.5 Postpartum interval

8.6 Pregnancy diagnosis.

# 9. Number of services/conception

All the 64 animals selected for the experiment were having history of multiple inseminations. The animals of group A were having inseminations ranging from 3–5 services per animal per conception whereas the animals of group B were having inseminations ranging from 2–4 services per animal per conception.

## 10. Milk production performance

Milk production of all the animals was also recorded. In group A, the average milk yield was 2 657 litters in 300 days per animal ranging from 1 836–3 064 litters in 300 days per animal. In group B, the average milk yield was 3 087 litters in 300 days per animal ranging from 2 190–3 532 litters in 300 days per animal.

# 11. Pregnancy diagnosis\_

Animals which did not return to estrus after 30 days post insemination were examined per rectum 60 days post insemination for the diagnosis of pregnancy.

**Tab. 1:** Comparison of age and lactation number in group A & B

Age and lactation	Group A (control)	Group B (treatment)
Average age (months)	83 ± 6.77	$87 \pm 5.42$
Range (age months)	48–232	44–186
Average lactation (NOS)	$2.44\pm0.38$	$2.47\pm0.44$
Range (lactation NOS)	1–13	1–11

Va	lues	between	groups	differ	non-significantly	/ (	$(\mathbf{P} > \mathbf{I})$	0.05	)
			0				-		

**Tab. 2:** Comparison of conception rate in group A (control) and B (Dalmarelin treated) cows 60 days post insemination

	Group A (control)	Group B (treatment)	Total
Number of animals	32	32	64
Animals pregnant	12	22	34
Animals not pregnant	20	10	30
Conception rate	37.5%	68.75%	53.12%

# 12. Data analysis

All observations were recorded on daily basis. Data was statistically analyzed by using Chi-Square tests (Agresti, 1996).

#### RESULTS

### Age and lactation number

In group A (control) the average age of 32 animals was 83 months  $\pm 6.77$  and age range was from 48–232 months. The average lactation number was 2.44  $\pm$  0.38 and the range number was from 1–13. Whereas, in group B (Treatment) the average age was 87 months  $\pm 5.42$  and range was 44 to 186 months. The average lactation number was 2.47  $\pm$  0.44. The lactation number ranged from 1–11 (Table 1).

Age and lactation number differ non-significantly (P > 0.05).

# **Conception rate**

In group A (control) out of 32 animals a total of 12 animals conceived and declared pregnant after 60 days post insemination, with conception rate of 37.5%. In group B (Treatment by an injection of Dalmarelin 2 ml at the time of artificial insemination) out of 32 animals 22 declared pregnant after 60 days of post insemination. The conception rate of group B remained 68.75% (Table 2). Comparison of conception rate revealed significant difference between group A and B ( $X^2 = 5.08$ , P = 0.02)

### **Postpartum interval**

The comparison of post partum interval between Group A and B are presented in Table 3. In group A (control) postpartum interval was  $528.5 \pm 55.35$  days and it ranged from 204–1 358 days whereas in group B (treated) postpartum interval was  $681.4 \pm 61.20$  days and it ranged from 219–1 494 days. This postpartum interval is before the start of treatment having no significance for comparison between the 2 groups (P > 0.05).

# Estrus duration and estrus behaviour\_

Table 4 illustrates the comparison of estrus duration and estrus behaviour in Group A (control) and B (Treated). The average estrus duration was 15 hours in group A and it ranged from 7–24 hours. Whereas in group B the estrus duration average was also 15 hours and range was 8–24 hours. Estrus duration differs non-significantly (X = 0.29, P > 0.05)

As for as estrus behaviour is concerned in group A (control), 13 animals showed intense estrus behaviour and the percentage was 40%, 14 animals showed mode-rate behaviour and the percentage was 44% and 5 animals showed weak estrus behaviour and the percentage was 16%. Whereas in group B (treated), 15 animals showed intense behaviour with percentage 47%, 13 animals showed moderate estrus behaviour with percentage 41% and only 4 animals showed weak estrus behaviour with percentage 12%.

The data on estrus duration and estrus behaviour revealed non significant difference ( $x^2 = 0.291$ , P > 0.05).

### DISCUSSION

The project was carried out to study the effect of Dalmarelin (GnRH analogue) on repeat breeding crossbred cows particularly in relation to improvement of conception rate, in these animals.

Tab. 3: Comparison of post partum interval in group A & B

	Group A (control)	Group B (treatment)
Postpartum interval (days)	528.5 ± 55.35	681.4 ± 61.20
Range (days)	204–1 358	219–1 494

Values between group A and B differ non-significantly (P > 0.05)

**Tab. 4:** Comparison of estrus duration and estrus behaviour in group A & B estrus duration

Estrus duration	Group A (control)	Group B (treatment)		
Average estrus duration (hours)	15	15		
Range (hours)	7–24	8–24		

Estrus behaviour

	Group A		Group 1	В
Estrus behaviour	control (No of animals)	% age	treatment (No of animals)	% age
Intense	13	40	15	47
Moderate	14	44	13	41
Weak	5	16	4	12

Values between group A and B differ non-significantly  $(X^2 = 0.291, P > 0.05)$ 

The Group B cows were given 2 ml of Dalmarelin at the time of AI. Upon rectal palpation 60 days following AI 22 out of 32 cows were found pregnant. Thus 68.7% conception rate was observed which is in accordance with the observations of Rangnekar et al. (2002) who reported 20 repeat breeders Holstein Friesian cows in estrus were divided into two groups. The first group (n = 10)was treated with 2.5 ml Fertagyl by intramuscular injection upon AI. In the treatment group, 7 cows conceived (70%). The improved conception rate in the treatment group may be due to the beneficial effect of Fertagyl in regulating the time of ovulation in cows. The similar observation was reported by Shelar et al. (2002). In the treatment group, 10 cows were inseminated at standing estrus and 2.5 ml receptal (10 ug buserelin) was injected intramuscularly at the time of insemination. The conception rate of the GnRH group was 60%.

The females with postpartum interval 528.5 + 55.35 days (range 204–1 358 days) in group A and 681.40 + 61.20 days (range 219–1 494 days) in group B were used in this study. Similar experiment was conducted by Kishore and Toshihiko (2006) who selected repeat breeding buffaloes with 12 months or more postpartum interval. They treated them with either GnRH or PGF 2  $\alpha$  and showed a satisfactory conception rate after treatment.

The average duration of estrus observed in group A was 15 hours with a range of 7–24 hours. While in group B, average estrus duration was 15 hours with an average of 8–24 hours. These findings are in agreement with the observations of Kanai and Shimizu (1982) who reported average duration of estrus 17.3 + 4.6 hours (range from 9–24 hours). In group A, 40%, 44% and 16 % females showed intense, moderate and weak estrus behaviour respectively. In group B, 47%, 41% and 12% females exhibited intense, moderate and weak estrus behaviour respectively. These results are in accordance with the work of Morales et al. (1983) who found intense, moderate and weak estrus behaviour ate and weak estrus behaviour in crossbred cows.

In group A, out of 12 pregnant animals, 8 were found from age between 48–70 months. Whereas in group B, out of 22 pregnant animals, 14 were found from age between 71–100 months. This study showed that repeat in breeding crossbred cows between 71–100 months age responded well to GnRH treatment. In this neither regard we cannot compare our results with other studies as neither such study has been reported nor are comparable references available.

Out of 12 pregnant animals in group A, 10 were seen from lactation number 1–3. Whereas out of 22 pregnant animals in group B, 19 were seen from lactation number 1–3. It was revealed that repeat breeding crossbred cows in their lactation number 1–3 were very well responsive to GnRH treatment. In this context no comparable references were available. In conclusion the findings of our study revealed that use of GnRH analogue therapy improved the conception rate in repeat breeding cross breed cows, when it was Administered at the time of AI. Moreover repeat breeding crossbred cows in 1-3 lactation are more responsive to GnRH treatment. It is therefore suggested that GnRH analogue would be effectively used to overcome the repeat breeding in crossbreed cattle.

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Received for publication on January 7, 2009 Accepted for publication on October 19, 2009

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